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STATE COLLEGE OF WASHINGTON
AGRICULTURAL EXPERIMENT STATION
Pullman, Washington

Division of Plant Pathology

The Rots of Washington Apples in Cold Storage

by

F. D. Heald and G. D. Ruehle

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(2). Of this list of 34 troubles, only eight are of sufficient importance to demand the attention of orchardists in the central irrigated valleys, while one-half of the prevalent diseases do not directly affect the fruit. It may be noted that such troubles as scab, bitter rot, black rot, blotch, rust, fruit spot and sooty blotch, all fungous diseases which mar, deform or destroy fruits by their direct attack are either absent from our commercial districts or, in one or two cases, are present in traces only, and therefore, are of little or no commercial importance. It is a recognized fact that the apple crop from the central irrigated valleys is practically free from either decay or other fungous lesions at the time of harvest.

Classification of Fruit Defects

The fruit defects which lower quality in our Washington apples or exclude them from the market, may be classified as follows:

(1) *Fungous defects,* comprising storage rots, mildew russet and scab which is not present in our commercial districts; (2) *Insect defects,* outstanding examples being codling moth, San José scale, aphis, etc.; (3) *Physiological defects,* including bitter pit, cork, drought spot, internal breakdown, Jonathan spot and freckle, scald, water core, etc.; (4) *Chemical injuries* from ammonia, arsenic, hydrochloric acid, sulphur dioxide, box wood, sprays, etc.; (5) *Climatic injuries* such as freezing or frost injury, sun burn, etc.; and (6) *Mechanical injuries* including stem punctures, box bruises, limb rub, pulled stems, and various other skin blemishes or defects. Certain of these defects may be initiated previous to harvest and be in evidence at packing, others may be initiated during the harvesting and packing operations, while others may develop only during storage, transit to market, or while awaiting consumption.

A large proportion of the crop each year must be stored for longer or shorter intervals before it can be marketed. It is during this storage period when the fruit is held in either cold or common storage or during transportation to market, that most of the loss from fruit decay is sustained. It is of importance to recognize the various types of fruit defects, since many of them have a very important bearing on the incidence and development of fungous decay which will be discussed in the following pages.

Importance of Decay in Storage or During Transit to Market

A real conception of the importance of the decay problem can only be formed by following fruit through storage to the ultimate consumer. No data are available to represent the actual total loss, but records of inspections for condition at various times during storage and the car-lot inspections at eastern terminals are illuminating. Some figures from condition reports for 1929 may be noted,[1] the emphasis being placed on blue mold decay. Many car-lots have shown 2 to 3 per cent of decay, a considerable number 3 to 7 per cent decay, a smaller number 8 to 10 per cent, while some few lots have shown from 12 to as high as 40 per cent of decay, necessitating either repacking previous to shipment or reduced prices at destination. The inspections of car lots at eastern terminals are even more convincing of the heavy losses sustained (21). Apparently many cars move to market from storage without inspection, otherwise it would be difficult to explain why the percentages of decay are frequently much higher at eastern terminals than for fruit held in local storage. Frequently one-third or more of the car lots show decay with some car averages as high as 50 or more per cent with box lots in certain cars as high as 75 to 90 per cent decay. Each year, on terminal markets, a considerable number of cars of apples are rejected by buyers for various reasons. In a study of the causes of such rejections of Washington apples in eastern markets for 1922-25, it has been reported (39) that an average of 21.3 per cent of such rejections were due to decay alone.

Further data will be presented later in a special discussion of the blue mold problem, but the facts here presented should be sufficient to emphasize the importance of the losses to the apple industry caused by storage decay. No careful and detailed study of the organisms responsible for the decay of Washington apples has been made previous to the study on which this report is based. During the past four years, special attention has been given to the fungi causing decay under cold storage conditions, working in the main with the more important commercial varieties from the central irrigated valleys.

[1] Figures obtained from office of Horticultural Inspector, Wenatchee, Washington.

TYPES OF APPLE ROTS IN COLD STORAGE IN WASHINGTON

The decay of apples in storage is due to the inroads of rot-producing fungi many of which can gain entrance into sound fruits only through mechanical injuries or necrotic spots, while a few are capable of penetrating the unbroken epidermis. Bacteria are rarely responsible for storage decay, the acidity of the cell sap being in general unfavorable for bacterial growth. A survey of the literature from various countries reveals a very large list of fungi which have been reported as causing decay in apples, the compilation totaling 90 species. The study made during the past four years has revealed more than 40 species, belonging to 22 genera, the record being based on a systematic examination of 5 to 10 box lots of the various varieties, with the necessity of a large number of isolation cultures and microscopic examinations.

In previous discussions of storage rots of the Pacific Northwest, four main groups (21) have been recognized: (1) *Blue mold decay* caused by several distinct species of Penicillium, of which one, *P. expansum* is of outstanding importance; (2) Gray mold rots, caused by several species of unrelated fungi, which generally show one character in common, a grayish or whitish growth whenever the fungi come to the surface of the rotted areas; (3) Black rots, generally referred to by the Inspection Service in the Pacific Northwest as *Alternaria rots* and characterized by the dark brown or black coloration of the decayed tissue, which is generally much firmer than in the blue or gray mold rots, and (4) *Anthracnoses* represented by the fruit-rotting phases of the black-spot or Pacific Coast canker, and the perennial canker, the latter under the name of false anthracnose or bull's-eye rot. In the following discussion this classification will be followed in part only, the emphasis being placed on generic groups of the causal organisms, rather than on such artifical characters as color. Since this grouping is to be followed, a list of the fungi found to be connected with the decay of Washington apples in cold storage will first be presented.

The following fungi have been isolated and proved by inoculation tests to be capable of causing decay either at cold storage or at higher temperatures:

PHYCOMYCETES

Mucor piriformis Fischer
Rhizopus nigricans Ehr.

ASCOMYCETES

Pleospora fructicola (Newton) Ruehle
Mycosphaerella tulasnei Jancz.

FUNGI IMPERFECTI

Phoma, No. 1
Phoma, No. 2
Coniothyrium, No. 1
Coniothyrium, No. 2
Microdiplodia, sp. undet.
Gloeosporium perennans Z. & C.
Pestalozzia hartigii Tub.
Coryneum foliicolum Fück.
Oospora, sp. undet.
Cephalosporium carpogenum n. sp.
Penicillium expansum Lk.
" *puberulum* Banier
" *verrucosum* Biourge
" *olivino-viride Biourge*
" *viridicatum* Westling
" *martensii* Biourge, and five other unidentified species.
Sporotrichum malorum Kidd & Beaum.
" *carpogenum* n. sp.
Botrytis cinerea Pers.
" *mali*, n. sp.
Cladosporium malorum n. sp.
" *herbarum* Lk. (See *Mycosphaerella tulasnei*)
Hormodendron cladosporioides (Fr) Sacc.
Stemphylium congestum Newton
" " Newton, var. *minor* Ruehle.
" (See *Pleospora fructicola*)
Alternaria tenuis Nees.
" *mali* Roberts
" No. 3
" No. 4
" No. 5
Fusarium No. 1
Fusarium No. 2
Ramularia magnusiana (Sacc.) Lind.
" No. 2
Epicoccum granulatum Penz.

BASIDIOMYCETES

Corticium centrifugum (Lev.) Bres.

Penicillium or Blue Mold Decay

Every worker who has dealt with the cause of decay in apples has reported one or more species of Penicillium. The earlier workers were inclined to refer any blue mold attacking apples to *P. glaucum* Lk. and

this name has been retained by some writers even to the present time. It has been shown that this name was too loosely used and a species-aggregate of strains and varieties grading into each other is now recognized as *P. expansum* Lk., the most common and destructive of the various species attacking apples. (Fig. 10A) The *P. expansum* type of blue mold has been the form most frequently met in our storage studies, but a number of other less common species, generally also less active agents of decay have been encountered.

It has been estimated that blue molds cause 80 to 95 per cent of the storage decay of apples (6, 47). These figures will not hold for the apple crop of the Northwest produced in localities where perennial canker rot or bull's-eye rot is especially prevalent, but it is conservative to say that blue mold is responsible for at least 75 per cent of the storage rot in apples from other localities in Washington.

The appearance of the decay caused by most of the species of Penicillium is very similar, the rotted areas being soft and watery, light brown in color, and in lesions of any considerable age, accompanied by the surface production of a bluish-green sporulating growth, which is nearly snow white in its initial stages. The lesions are of varying shades of brown, being lighter on the yellow or green varieties and on the pale cheek of colored varieties and showing the darker shades of brown on the most highly colored varieties. The soft watery consistency of the rotted tissue seems to be a more distinguishing feature than the color variations. Two characters are of importance in the recognition of *P. expansum,* the most common species, namely, the musty odor and the formation of conidial tufts or coremia (Fig. 1A) on the surface of well-developed lesions. Some of the other species may produce a heavy surface sporulation but without the formation of the characteristic spore tufts. This description holds true for all of the species studied except for certain species of the *P. viridicatum* series. In these species the lesions are generally a darker shade of brown, the decayed tissue firmer, always lacking a surface growth under cold storage temperatures, and very slow-growing (Table 1, Nos. 7, 8 and 9) as compared with *P. expansum* (Figs. 2 and 3).

From examinations made early in the storage period of the various lots of apples observed it was found that approximately 80 per cent of the rot lesions were of the blue mold type. Later examinations of the same

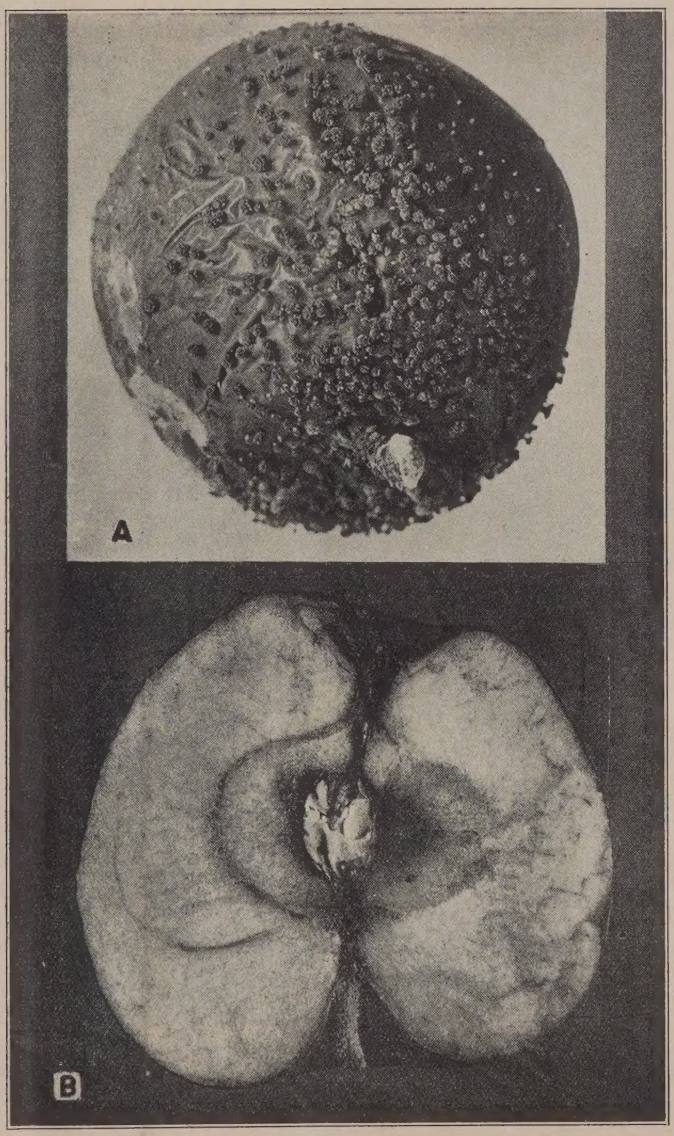

Fig. 1. A, Complete decay caused by blue mold, **Penicillium expansum**; B, Moldy core of Stayman Winesap.

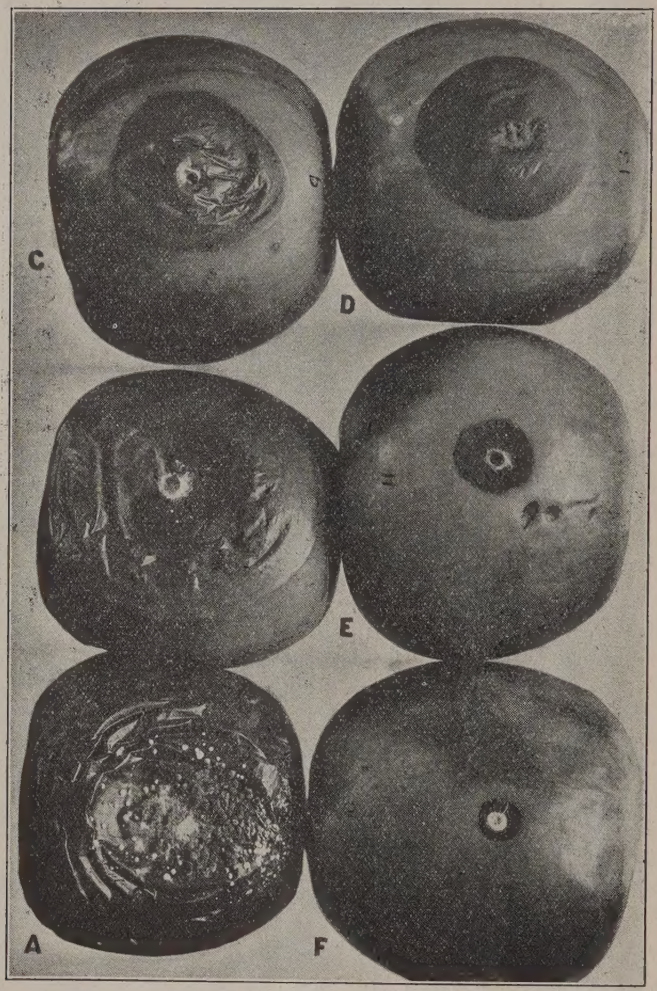

Fig. 2. Jonathan apples from cold storage 94 days after inoculation with Penicillium species. A, B, P. expansum; C, Penicillium No. 9; D, P. martensii; E, P. olivino-viride; F, P. verrucosum.

lots reduced the total percentage of blue mold lesions for the entire season to less than 50 per cent, since many slow-growing fungi were found to occur in late storage. Many of the late examinations were made, however, after the ordinary commercial limit for storage of the varieties in question, so that the early examinations give a truer estimate of the actual commercial losses.

All of the blue molds are primarily wound parasites, most frequently gaining entrance through fresh mechanical injuries such as stem punctures, bruises, insect injuries or necrotic tissue of diverse origin, but sometimes through normal stems or open calyx canals. The lesions may be on the check, at either stem end or calyx end, or as a core rot, originating from mold that has penetrated an open calyx canal. In ordinary commercial stock a single lesion to a fruit is the common condition. Under cold storage conditions blue mold lesions caused by the common species may be expected to be one to one and one-quarter inches in diameter at the end of eight to ten weeks.

The definite recognition of the different species of blue mold is only possible by means of laboratory cultures and microscopic examination, and even then their positive determination is difficult because of the slight differences. For present purposes no attempt will be made to present diagnostic characters, but that the various species differ in their ability to produce decay may be illustrated by the results of inoculations recorded in Table 1.

Table 1. Rate of decay produced on ripe Jonathan apples by various strains or species of blue mold isolated from rotting apples.

Name or number of culture	Range in diameter of lesions in millimeters		
	After 12 days at 68-77° F.	After 84 days in common storage at 41-50° F.	After 94 days in cold storage 30-35° F.
No. 1 *P. expansum*	50-60	60-65	60-65
No. 2	34-45	55-60	32-40
No. 3	13-16	15-20	30-35
No. 4	12-15	15-20	30-38
No. 5	8-15	18-30	30-35
No. 6	20-36	15-20	10-25
No. 7	6- 9	10-17	7- 8
No. 8	6- 9	6- 9	9-15
No. 9	10-15	9-17	7- 8

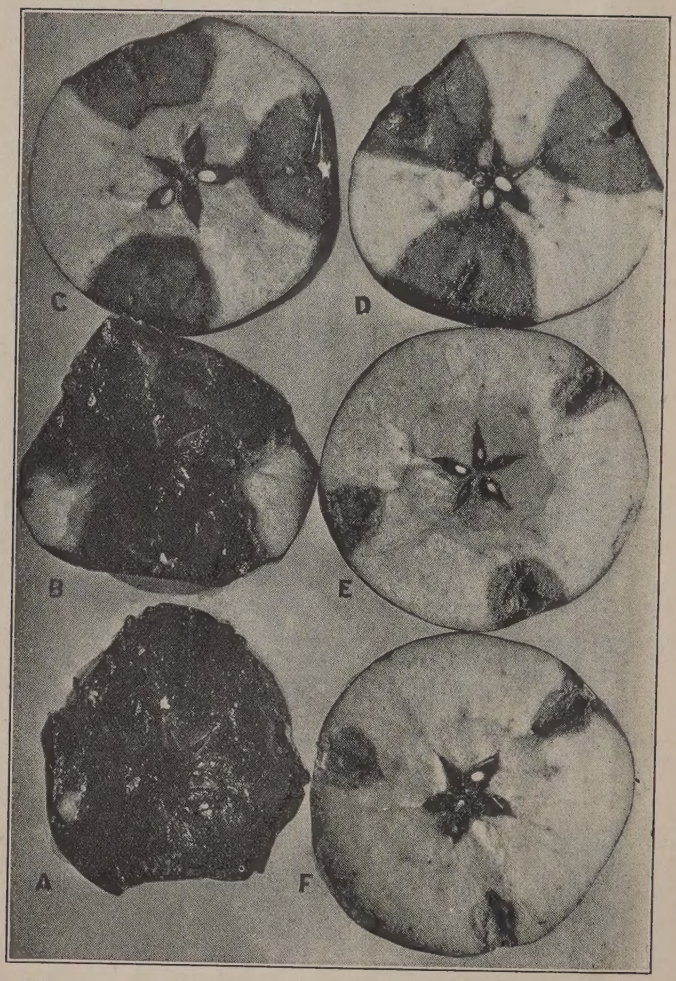

Fig. 3. Transverse sections through the apples shown in Fig. 2 showing the extent of penetration of the decay by different Penicillium species.

It may be noted that the common species, *P. expansum* is able to rot apple tissue almost twice as rapidly as any of the other isolations, and about nine times more rapidly than the slowest-growing species. *P. expansum* not only exceeds the other species in its rate of advance in apple tissue, but also in its frequency of occurrence, so that it far overshadows all other species as a cause of storage decay. The importance of the other species should not be ignored, since, it seems probable that under certain conditions the attack of these less frequently occurring forms may assume considerable importance. Futher discussion of the blue mold problem is reserved for later consideration.

Gloeosporium Rots

Two species of Gloeosporium have been reported from Europe as capable of causing serious rotting of apples: *G. album* Osterw. and *G. fructigenum* Berk. (30, 45), the former causing a rot apparently very similar to that produced by *G. perennans*, Z. & C., a species peculiar to the Pacific Northwest; the latter considered to be forms or strains of the conidial stage of the bitter rot fungus, *Glomerella cingulata* (S.) S. & von S. In our study of Washington apples, two species have been encountered, the fruit-rotting or Gloeosporium phase of the black-spot canker or Northwestern anthracnose, *Neofabraea malicorticis* (Cord.) Jackson, and the fruit-rotting phase of the perennial canker, *G. perennans* (Fig. 6A), frequently referred to as bull's-eye rot (13, 14, 48). The former does not occur in the main commercial apple districts, so will be ignored in the

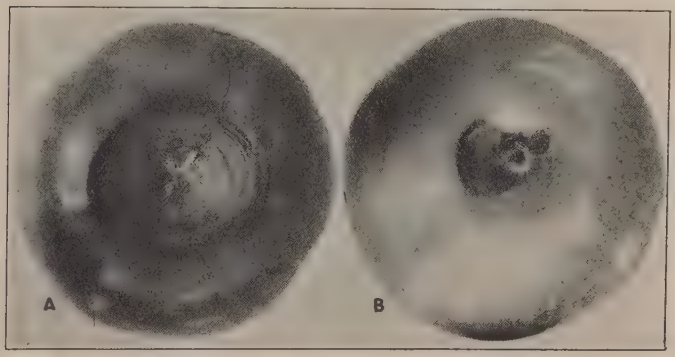

Fig. 4. A. Stem-end decay caused by **Gloeosporium perennans**; B. Calyx-end decay caused by **Gloeosporium perennans**.

present consideration. Perennial canker or bull's-eye rot is a serious storage trouble of apples in the White Salmon section and adjacent Oregon, occurs sparingly in the Wenatchee and Yakima valleys, and may occasionally be found on apples from any of the other districts of Eastern Washington.

In its most typical form the bull's-eye rot appears in late storage as small, circular, slightly sunken, fairly firm lesions, which are either light or dark brown in color, and show a lighter brown or tan center surrounded by a darker zone of brown (Fig. 5A). It is this concentric zonation which has suggested the common name of bull's-eye rot.[1] In the younger lesions, the fungus does not apper on the surface, but where older or more extended, spore fruits or acervuli break through and produce creamy white or grayish gelatinous masses of spores. The abundance of the spore-fruits is increased by humid storage and when held in a moist atmosphere there will be some surface development of mycelium in tufts or in less aggregated form. The lesions may be few in number on a given fruit or very numerous, and may occur at any place on the surface. In severe spotting the lesions appear to center at lenticels, but they may originate readily from any form of mechanical injury. Early infections frequently originate around the stem in the basin or at the calyx end (Fig. 4). The color and consistency of the rotted tissue and the type of surface growth serve as a ready means of recognition in contrast to similarly located lesions of blue mold.

In regions in which the perennial canker fungus is generally established on the trees, the bull's-eye rot, is of even more importance than blue mold, especially in favorable seasons. The serious aspect in this rot is the fact that infection is not confined to mechanical injuries, since the fungus is able to enter through the lenticels. If the inoculum of spores is present, perfectly sound apples may develop numerous rot lesions under either cold or common storage. The extent to which the bull's-eye rot will develop in storage is influenced by the conditions which prevail previous to harvest, and is also favored by high humidity during storage. If the fruit is harvested, cleaned and packed before rains of any consequence, the amount of bull's-eye rot will be reduced to a minimum, for that par-

[1] In addition to the perennial canker fungus, five other fungi produce the bull's eye type of decay. Heald, F. D. Bull's-Eye Rots of Apples in Washington. The Wenoka Arrowhead 3: 1. 6. Mar., 1931.

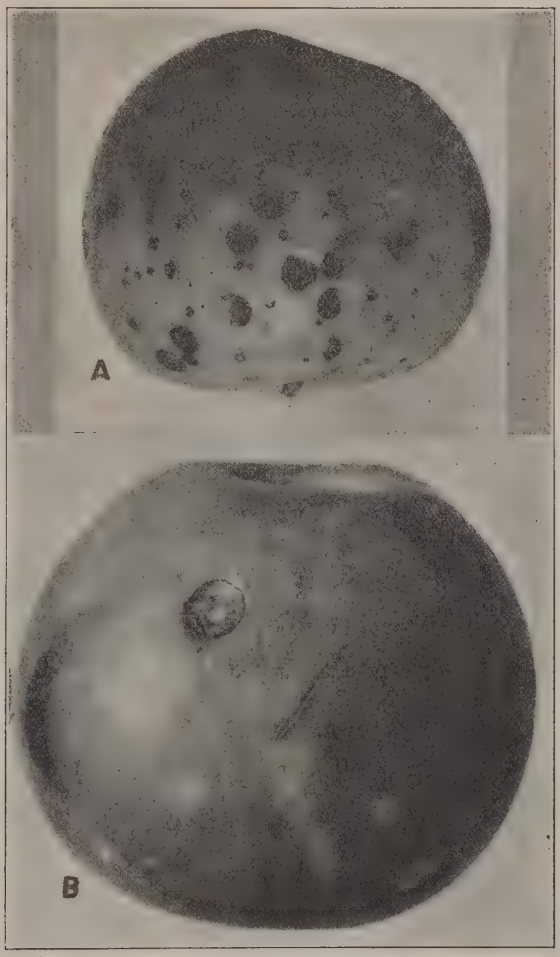

Fig. 5. A, Bull's-eye type of spot rot caused by **Gloeosporium perennans**; B, Dark slow-growing spot rot caused by **Pleospora fructicola**.

ticular environment or set of storage conditions, while with a rainy harvest season the rot may be expected to be severe. The basis of this behavior is that the spores causing the infection originate from cankers on the trees, and are not wind-disseminated but are readily washed down by rains and so may be present on the fruit at time of picking. This relation was well illustrated by the severe development of the rot in apples from the White Salmon section in the crop of 1927, when some individual boxes showed as high as 60 per cent infection in February, while in 1928, which was a dry fall, the amount of decay was very moderate. A study of the orchards in the White Salmon section has shown a decreasing abundance of sporulating cankers, with a small number of new infections, so that there is not now available the abundance of inoculum that was present a few years ago.

Our experience with the rot has been largely with the Jonathan, Yellow Newtown and Spitzenberg varieties from the White Salmon district, and these appear to show increasing susceptibility in the order listed, but the trouble has been encountered on Romes, Winesaps, and Delicious from other locations. How much of this variation is caused by real resistence and how much to the varying prevalence of the inoculum is somewhat uncertain.

Fig. 6. Isolation cultures on 2 per cent dextrose potato agar; A, **Gloeosporium perennans**; B, Oospora spp.

When inoculated into ripe apples of several varieties the fungus readily produced typical lesions, no marked varietal differences being manifested. At cold storage temperatures, the rotted areas measured 30-45 mm. in diameter after four months. At 68° F., the lesions developed slowly, requiring several weeks to reach noticeable size but they developed rapidly after one month. Bull's-eye rot must be regarded as one of the most serious types of decay of apples in the late storage of fruit that has been grown in orchards in which perennial canker is prevalent.

Alternaria Rots

Four distinct species of Alternaria have been shown to cause a decay of apples. Two of these species, *Alternaria pomicola* Horne and *A. grossulariae* Jancz., occur on apple fruit in England, but have not, as yet been reported on this host in America (25, 26). A third species, *A. mali* Roberts, in addition to causing enlargements of dead spots on apple foli-

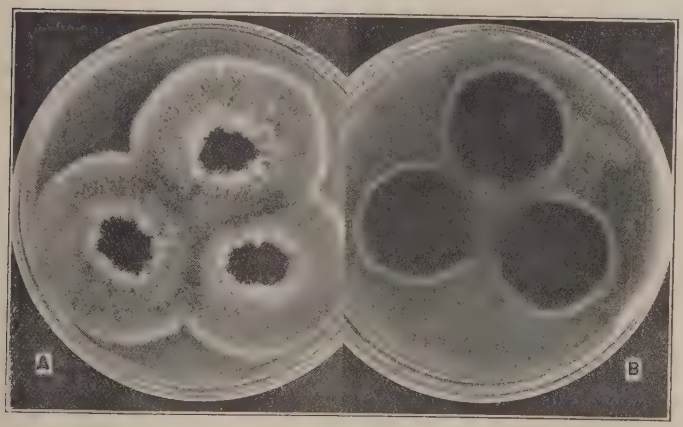

Fig. 7. Isolation cultures on 2 per cent dextrose potato agar: A, **Alternaria tenuis**; B, **Alternaria mali**, dark strain.

age, has been reported as capable of causing a fairly rapid rot of the fruit in America (40, 41). The fourth species, *A. tenuis* Nees, has been reported from both Europe and America as the cause of apple decay (30, 33). Alternaria rot is widespread and common, and since its discov-

ery by Longyear in Colorado in 1905, has been reported from nearly every country where apples are of commercial importance. As the result of recent studies, *A. tenuis* and *A. mali* were found to occur commonly and to produce a very similar type of decay on stored apples in Washington. The former produces fluffy colonies and large spores on culture media, and the latter forms colonies with scant aerial hyphae and somewhat smaller spores (Fig. 7). In addition, several other species or strains of Alternaria were found to occur infrequently on the fruit.

The lesions produced by Alternaria are invariably dark brown to black in color and are generally found to be centered around a break of some sort in the skin. Occasionally the rot may develop in the calyx, the basin or in soft scald areas, and the fungus has been frequently found in core rots. Jonathan and Rome Beauty apples seem to be more susceptible to Alternaria rot than Winesap and Delicious apples.

The dark lesions develop quite rapidly on ripe apples at 77° F., in some cases involving the entire fruit in two months. The affected host tissue may be rather soft and moist and as the rot enlarges, small black pimples may appear on the surface from the presence of minute black hyphal aggregates located just beneath the epidermis. Occasionally under moist conditions, the dark colored mycelium appears on the surface and produces the characteristic spores of the fungus.

The development of Alternaria rot is greatly delayed by cold. At cold storage temperatures, the lesions produced may reach 25 mm. in diameter, five months after inoculation, but usually they are smaller. The injury is more of the nature of black, shallow, dry spots than of active rots. Usually a cavity is formed beneath the epidermis, which is filled more or less completely with the dark colored hyphae of the fungus mingled with fragments of dead host cells. When apples with such spots are removed from cold storage to higher temperatures, the fungus may resume activity and the decay progress rapidly. From the nature of the injury at cold storage temperatures, it is apparent that Alternaria rot will not be a serious factor in apples that are stored at such temperatures very soon after picking.

Pleospora and Stemphylium Rot

A Pleospora rot of apples has been known in England since 1920 (24) and two species, *Pleospora pomorum* Horne and *P. herbarum* Pers. are

the causal agents. In 1927, a Pleospora rot of lemons from California and of apples from Washington was reported (41a) and the organism was identified as a variety of *P. herbarum*. Recently, Newton (35) described a Pleospora causing a decay of apples in Washington, which is distinct from the other species reported from apples. The name *P. mali* Newton was given to this species, but since this name had been previously preempted for the perfect stage of *Hendersonia mali* (23), the name *P. fructicola* n. comb. has been recently proposed for the fungus described by Newton (44).

All of these forms possess an imperfect or conidial stage, which may be classified as Stemphylium, in addition to the ascomycetous or Pleospora stage. In addition, two species of Stemphylium, for which the perfect or ascomycetous stage has not been discovered, are known to produce a decay of apples. The first of these, *Stemphylium graminis* (Corda) Bon. is reported on apple fruit from England, (30) and the second, *S. congestum Newton*, has been reported only from Washington apples (35). Recently a small-spored variety of *S. congestum* was described (42).

The lesions produced by Pleospora and Stemphylium are dark brown to black, fairly firm, and are indistinguishable from rots produced by Alternaria, without careful microscopical examination or culturing the fungus. In some cases, *S. congestum* produces its characteristic spores in the affected apple tissue and can then be readily identified by means of a microscopic examination of decayed tissue. Spores are not produced in every case, however, and are not present in the young lesions, so that it is usually necessary to culture the fungus for positive identification. *Pleospora fructicola* does not fruit on the apple, although immature perithecia are frequently formed just beneath the epidermis or deeply embedded in the fruit. The presence of these bodies gives a pimpled appearance to the surface of the cuticle, which then closely resembles the appearance of many Alternaria lesions.

There is not much difference in the rate of decay caused by Pleospora and Stemphylium. Pleospora invades apple tissue slightly faster at cold storage temperatures, but at higher temperatures the two fungi proceed at nearly equal rates. When compared to *Alternaria tenuis*, the latter was found to produce a more rapid destruction of apples at temper-

atures of 60° F. and 77° F., but at cold storage temperatures, the difference in rate of decay is not marked.

Pleospora fructicola is fairly common on cold storage apples in Washington, occurring about as frequently as Alternaria species and much more frequently than *Stemphylium congestum* (Fig. 11, B). Neither species assumes much importance in cold storage, since cold greatly inhibits their development. Under common storage conditions, however, the rot produced by these forms may be of considerable importance. No doubt a large percentage of decay attributed to Alternaria in the Northwest is due either to *Pleospora fructicola* or *Stemphylium congestum.*

Sporotrichum Rot

A fungus known as *Sporotrichum malorum* Kidd & Beaum. was first isolated from apples in storage in England in the winters of 1921-1922 and 1922-1923 (30). In 1928, Gardner (17) reported this species causing a shallow surface rotting of apples in Indiana, and in 1929 (18) published a complete account of the disease as it occurs in that state. The disease, which was described as "Sporotrichum fruit spot and surface rot" was reported as quite serious on Grimes, Winesap, and Ben Davis apples grown in southern Indiana. The symptoms of the Delicious spot disease, a storage rot described by Cunningham (10) in New Zealand, closely resemble the symptoms of Sporotrichum fruit spot, but no description of the causal fungus was given.

Sporotrichum rot was first noted in Washington in 1926, on some apples grown in the Kennewick district. The fungus was found to be morphologically identical with *Sporotrichum malorum*, a culture of which was obtained from Holland for comparison. In 1927 and 1928, the same species was obtained frequently from late storage rots developing on apples from Wenatchee, and more recently from the Yakima district. The disease was also found occurring on apples from White Salmon.

Sporotrichum lesions vary in color from light brown to dark brown, with sometimes a light brown area surrounded by a darker border, thus giving somewhat the appearance of decay caused by the perennial canker fungus. The rotted areas are usually rather shallow, and but slightly depressed, or they may extend deeply into the flesh in some cases. The decayed tissue is generally soft, and somewhat watery, and separates readily from the healthy tissue, but in some cases, under conditions not well

understood, may be dry and fairly firm. In either case, the characteristic mycelium and spores of the fungus are abundant in the affected parenchyma tissue of the apple. Under ordinary storage conditions, the fungus does not appear on the surface of the lesions. Entrance is effected either by way of mechanical injuries or necrotic areas in the skin or through lenticels.

The Sporotrichum was isolated generally from late storage rots, and from apples that were in the ripe stage of maturity. When fresh punctures were inoculated with spores of the fungus, infection readily took place on several varieties. The lesions develop very slowly at cold storage temperatures, and usually several months are required before they attain measurable size. An incubation period of five to six months is necessary for the formation of lesions 30 to 40 mm. in diameter. At higher temperatures (68° F.) the fungus produces decay at a faster rate, but is much slower in this respect than Alternaria or Pleospora. Just when natural infection occurs and what are the sources of natural infection are not known.

A second species of Sporotrichum, isolated from Washington apples, has been described as a new species, *Sporotrichum carpogenum* (44). It is capable of producing a decay of the same type as that produced by *S. malorum*, but is much less common than the latter.

Botrytis or Gray Mold Rot

Three species of Botrytis have been reported as occurring on the fruit of the apple (38). One of these, *Botrytis ramosa* Pers., is so imperfectly described that it is impossible to be sure of its validity, but the others have been shown to be synonyms of the common and widespread *B. cinerea* Pers. (Fig. 8B). The latter species is apparently universal in its distribution and occurs on many different hosts. It has been reported as causing a storage rot of apples from various points in Europe, Australia and North America (1, 7, 22, 45). It has long been known to cause decay of apples in Washington, and occasionally causes considerable damage, especially in the more humid districts of the state.

Botrytis lesions are generally light brown in color. At first they have a pale, translucent, watery appearance which soon changes to a dull brown. Young lesions have much the same appearance as those produced by *Penicillium expansum* Lk. but they are somewhat firmer and lack the

musty odor and taste so characteristic of the common blue mold rot. Under certain storage temperatures (*22*) many of the lenticels in the decayed areas are surrounded by a darker brown ring giving a polka-dot appearance which has suggested the name of "spot-rot" for this condition. At cold storage temperatures the gray mold has never been observed to sporulate on the apple, but frequently grayish tufts of sterile hyphae are pushed through breaks in the skin covering the lesion. Under moist conditions, at laboratory temperatures, conidial tufts soon appear and in a few days the fruit is a mass of gray mold, hence the name "gray mold rot" for this type of decay.

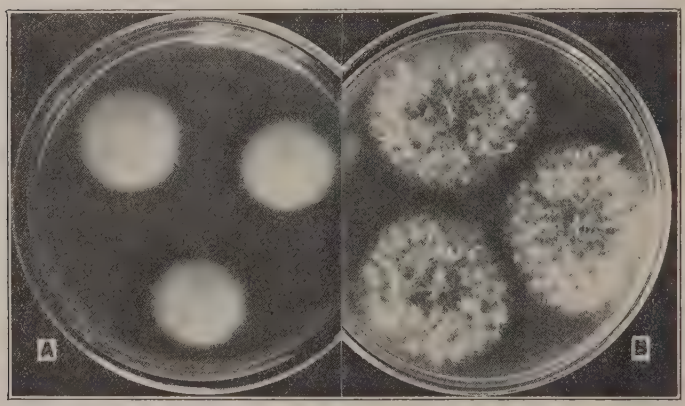

Fig. 8. Isolation cultures on 2 per cent dextrose potato agar; A, **Fusarium** spp.; B, **Botrytis cinerea.**

Botrytis has been repeatedly isolated from rot lesions occurring on several varieties of apples in cold storage but it is probably more frequent in common storage. Such lesions are usually well advanced, the entire fruit often being involved, and have generally been found fairly early in the storage season. In all cases observed, the decayed areas were found to have their origin either at breaks in the skin due to mechanical or insect injuries or at the calyx end of the apple. The fungus is apparently unable to penetrate the unbroken skin of the apple, since when a suspension of spores was sprayed on the surface of normal apples, no infections resulted. When fresh punctures were inoculated with spores, infection readily took place. At 77° F. the fungus produced a very rapid

decay of ripe apples, completely destroying the fruit in 20 days. At 60° F., the decay was much slower, (Table 2), the lesions reaching 2-3½ inches in diameter at the end of 14 days. In cold storage the rate of progress was still slower, but frequently ripe Jonathans were over half to three-fourths decayed at the end of a 60-day incubation period. The spot-rot symptom developed only in lots held at 60° F. The inability of the fungus to fruit on the apple at cold storage temperatures, renders its chance of spread in cold storage slight. Inoculation probably takes place before or during the packing process, and infection may be established before the packed fruit reaches the cold rooms.

A second species of gray mold, *Botrytis mali*, has been described as a new species (44). This species is capable of producing a rapid decay of the same type as that caused by *B. cinerea*. It is much less common than *B. cinerea*, develops slightly slower at all temperatures and has not been observed to cause the characteristic lenticel spotting in the decayed areas.

Table 2. The Rate of Development of Gray Mold and Blue Mold, at 60° F. Innoculations Made December 6, 1929.

Variety	Gray Mold		Blue Mold	
	Average surface diameter of 12 lesions after 11 days	Average daily increment	Average surface diameter of 12 lesions after 11 days	Average daily increment
Jonathan	65.7 mm.	6.0 mm.	25.6 mm.	2.3 mm.
Delicious	67.8 mm.	6.2 mm.	34.0 mm.	3.1 mm.
Rome	62.7 mm.	5.7 mm.	29.5 mm.	2.7 mm.
Winesap	35.3 mm.	3.2 mm.	23.8 mm.	2.2 mm.
Stayman	53.6 mm.	4.8 mm.	29.1 mm.	2.6 mm.

The average daily increment of blue mold in cold storage was .5+ mm. Gray mold lesions advanced about two to three times as rapidly.

Mucor and Rhizopus Rots

Several species of Mucor have been reported from Europe as occurring on the fruit of the apple, but these appear to be referable to three species, *Mucor racemosus* Fres., *M. mucedo* L. and *M. piriformis* Fischer (38). There are but few reports of any Mucor rots of apples in America and

then only on market stock rather than in cold storage. In the present study a soft, watery, light brown rot, caused by *M. piriformis,* was encountered early in the season on Jonathan apples from Wenatchee. The infected fruits were completely rotted when boxes were taken from cold storage in December, the paper wraps being wet and sticky from the fluid draining from the rotting tissue. This oozing of liquid was frequently sufficient to extend to apples several layers below the infected ones. The fungus was found sporulating on the fruit, with its mycelium coming to the surface through lenticels and skin breaks, where numerous characteristic sporangial heads were produced.

Mucor piriformis is a very rapid grower and is able to cause a rapid decay at cold storage temperatures or higher. Under cold storage conditions lesions were 20-25 mm. in diameter at the end of one month and after a lapse of two months a large percentage of the inoculated apples was completely decayed. It may be noted that this was a more rapid decay than that caused by the common blue mold (See Table 2). At 60° F. inoculated fruits were completely rotted at the end of ten days to two weeks, while at 77° F., the decay started very quickly but progressed slowly and sometimes stopped entirely after lesions 20-30 mm. in diameter had been produced. It is rather surprising that this rot is not more common on cold storage apples, since it exceeds blue mold in its rate of development and also sporulates freely on fruit at temperatures close to 32° F. It seems likely that the spores are much less numerous and less tolerant of adverse conditions than the common blue mold. While this fungus is an infrequent cause of decay in apples, the potential possibilities of causing heavy losses under certain favorable conditions should be recognized.

Another rot of apples known as Rhizopus rot is practically indistinguishable from that caused by *Mucor piriformis.* This rot, caused by *Rhizopus nigricans* Ehr., has been reported by investigators in widely separated parts of the world, in Europe, Australia and America, (1, 7, 30, 34, 45) but it has never been observed to cause considerable loss to apples early in the season. The fungus has never been observed to form a surface growth of fruiting hyphae on apples in cold storage, the mycelium remaining internal, but at higher temperatures, and especially under moist conditions, the mycelium appears through the lenticels or breaks in the skin and produces numerous fruiting branches or sporangiophores.

In inoculation experiments it was shown that our isolation of Rhizopus was unable to produce decay in ripe Jonathan apples held under cold storage conditions, but apples were completely rotted at 60° F. in three or four weeks' time, and at 77° F. in about one week. It seems probable that the few cases of Rhizopus rot appearing in cold storage were the result of infections established before the fruit reached cold storage.

Rhizopus rot, like Mucor rot, must be considered a very minor type of storage decay, most likely to appear in common storage or on the market.

Cladosporium Rots

Cladosporium species have been mentioned by various workers as associated with apple rots. *Cladosporium herbarum* Lk. was reported by Morse and Lewis (34) as able to make a slight growth in the apple, by Schneider-Orelli (45) as causing small black spots in certain varieties, and by Kidd and Beaumont (30) as capable of direct infection of the apple, but the latter concluded that this fungus was of but little consequence as an independent parasite of apples. Another species referred to *C. epiphyllum* Pers. has been repeatedly isolated from apple spots in England (26) but later workers have considered it indistinguishable from the common *C. herbarum*.

The spores of Cladosporium are more abundant on the surface of normal sound apples than any other species of fungi, being much more numerous than the species of blue molds. The mycelium is dark and always causes a darkening of the tissue which is invaded, lesions of any age being frequently nearly coal black. Cladosporium species frequently invade scalded tissue and contribute very largely to the black colorations that are so characteristic of severe types of storage scald. They also play a prominent part in the production of the dark brown or black margins around worm holes or mechanical injuries not invaded by the more actively growing fungi. In addition to these secondary effects it may be noted that fungi of the Cladosporium type have been repeatedly isolated from small, brown to black, firm, shallow lesions that developed on apples in cold storage. These lesions did not exceed 20 mm. in diameter and invariably occurred at bruises, stem punctures, insect injuries or other breaks in the skin of the fruit.

During the past four years, hundreds of isolations from dark lesions on apples have yielded fungi of the Cladosporium type. These isolations yielded eleven forms which were recognized as possessing fairly constant characters. Inoculation experiments with these species or strains gave only three that were capable of producing decay when inoculated into ripe Jonathan apples. These were as follows:

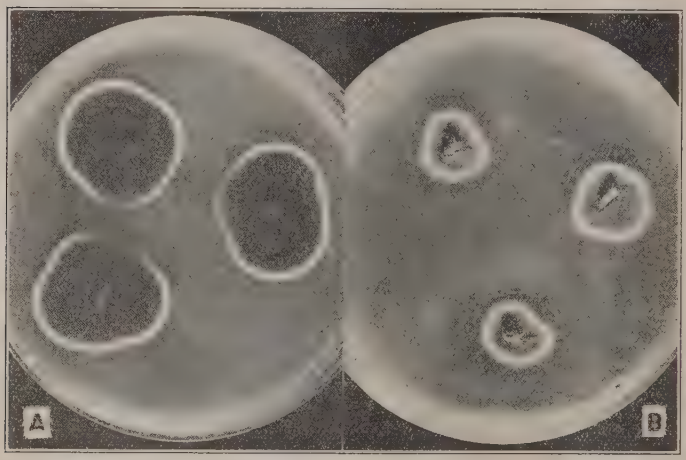

Fig. 9. Isolation cultures on 2 per cent dextrose potato agar; A, **Mycosphaerella tulasnei**, conidial or Cladosporium stage; B, **Hormodendron cladosporioides.**

1. The common species, *C. herbarum* (Fig. 9, A), which has been shown by this study (43, 44) to represent the conidial stage of the ascigerous fungus, *Mycosphaerella tulasnei* Janc., in confirmation of a claim first made in 1894 (29).

2. A closely related for (Fig. 9, B) generally referred to *Hormodendron cladosporioides* Sacc., which may represent a strain or stage of *C. herbarum*. It has constantly failed, however, to produce perithecia.

3. A new species of markedly different morphological and cultural characters that has been described as a new species, *C. malorum* (44).

Representative cultures of these types were grown on culture media with frequent transfers and at various temperatures for three years, and Type 2 was never observed to transform into Type 1.

Inoculation of cultures into sound apples showed that Types 1 and 2 produced lesions of nearly identical character and size. At cold storage temperatures, small, black lesions, 8-10 mm. in diameter were formed after three months, but these did not enlarge further after two more months of incubation. At 68°-77° F. ripe Jonathan apples developed small dark brown to black firm lesions which reached a maximum diameter of 25 mm. in three months although they were generally much smaller. Usually the affected tissue had shrunken, leaving a cavity under the skin filled with the dark-colored hyphae of the fungus.

The new species, *C. malorum*, was much more active in its growth when inoculated into sound ripe apples, as lesions 20-30 mm. were produced, in fourteen days at 68°-77° F., equalling or exceeding in size those produced by the other species at the end of three months. Although this species developed more rapidly at high temperatures, than the other two it made about the same advance in cold storage, the lesions not exceeding 10 mm. in diameter after five months.

Because of the slow growth of the causal fungi, Cladosporium spots appear mostly during the later months of the storage life of the various varieties, and rarely reach much size under cold storage conditions. Although not infrequent in late storage, the Cladosporium rots are of minor consequence in the storage life of apples.

Phoma, Coniothyrium and Microdiplodia Rots

The rots caused by species of Phoma, Coniothyrium and Microdiplodia will be considered in a group since the causal fungi are very closely related. All produce spores in pycnids, single-celled and hyaline in Phoma, single-celled and smoky or dark in Coniothyrium and two-celled, dark and under 15 microns in length in Microdiplodia. Several species of Phoma have been found to cause a decay of apples: *P. fuliginea* Kidd & Beaum. and *P. bismarkii* Kidd & Beaum. in England (30); *P. pomorum* Thüm. in Bulgaria (28); and *P. mali* Schulz. & Sacc. in the eastern United States (32). With few exceptions the attacks of these species seem of little importance. The rot caused by *P. mali* resembles that caused by black rot (*Physalospora cydoniae*) and spreads rapidly, usually with considerable white mycelium on the surface. The Polyopeus rots of England (24) are caused by species very closely related to Phoma.

Two distinct forms of Phoma recognizable by cultural and morphological characters have been isolated in this study from dark brown,

firm rots of apples. It has not been possible as yet to identify them as belonging to any of the species previously recorded from the apple, but for present purposes they may be designated as Phoma 1 and Phoma 2. The two species have shown a marked difference in their ability to rot sound apples, as may be noted in Table 3.

Table 3. Size of Rot Lesions of the Two Phoma Species When Inoculated into Ripe Jonathan Apples.

Phoma Number	Temperature 68° F.		Temperature 30-35° F.	
	Incubation period	Surface diameter of lesions	Incubation period	Surface diameter of lesions
No. 1	1 month	15-45 mm.	5 months	22-50 mm.
No. 2	1 month	7-10 mm.	5 months	- 0 mm.

Both species produced brown, fairly firm lesions, No. 1 with numerous pycnidia just under the epidermis, but No. 2 did not sporulate on the apple. No. 1 was isolated from Yellow Newtowns and No. 2 from Winesaps.

Several species of Coniothyrium have been reported as contributing to the decay of apples: *C. pirina* (Sacc.) Sheldon and *C. fuckelii* Sacc. in the United States (34, 36); and *C. cydoniae mali* Horne and *C. convolutum* Horne in England (30).

Two distinct species of Coniothyrium have been isolated in this study from dark brown, firm lesions. They have much larger spores than any of the species previously recorded from the apple, and may represent new species, but for present purposes they will be designated as Coniothyrium No. 1 and Coniothyrium No. 2. As in the two Phoma species, the two Coniothyriums have shown a marked difference in their ability to rot sound apples. Incubated at a temperature of 68° F., inoculations with No. 1 on ripe Jonathan apples developed lesions 50-60 mm. in diameter after two months, while No. 2 under similar conditions was a slower grower, making lesions only 15 to 25 mm. in diameter. Neither species was able to cause any rot when the inoculated apples were held in cold storage. Neither species produced pycnidia on the surface of the rotted tissue, but No. 1 produced them in abundance on the surface of sterilized apple wood or bark in cultures.

No records have been found of the occurrence of any species of Microdiploidia as causing decay in apples. A single species has been isolated a number of times from small, dark brown, firm lesions, never exceeding 20 mm. in diameter, essentially similar to those caused by other species in this group. This species was found capable of producing a brown, firm, shallow rot which developed slowly at temperatures of 68°-77° F., reaching a diameter not exceeding 25 mm. in two months' time.

Hypochnus or Fish-Eye Rot

A rot of apples caused by a species of Hypochnus was first reported in 1903 (12) as associated with and apparently following scab on Rhode Island Greenings and Baldwins in New York. Recently the increasing prevalence of an Hypochnus rot agreeing in essentials with the decay described from New York has been reported as quite common on New York Baldwins, and also on market stock of Jonathans from Idaho and Winesaps from Illinois and Washington (8). Later studies have shown that this causal fungus is *Corticium centrifugum* (Lev.) Bres. (9) and that certain sterile fungi producing the bull's-eye type of rots, should also be referred to this species. What appeared to be the same rot was observed to occur occasionally on Winesaps from the White Salmon district and from Wenatchee.

The appearance of the rot agrees in all essentials with that described by the other workers. Lesions produced either naturally or artificially generally show conspicuous tan centers, with a brown border, giving the characteristic bull's-eye effect so commonly found on apples affected with Northwestern anthracnose or perennial canker. The lesions are quite firm, smooth, slightly sunken, and circular except where two areas coalesce, but the necrotic tissue is somewhat tough and stringy in contrast to the soft consistency of the anthracnose lesions. A penetrating, sweet odor is given off from affected apples, and from cultures. The sweet odor, coupled with the presence of clamp connections in the mycelium (except in certain sterile strains) makes this rot and its causal fungus easy of identification. When an infected apple is placed in a moist chamber, a white mat of delcate, prostrate, non-sporulating, fungous filaments soon appears and radiates uniformly from the lesion. The Hypochnus rot appeared rather late in the storage season, either centering around lenticels or originating at the calyx.

Although the rot was found only on the Winesap of our storage stock, the fungus was found to be readily capable of infecting Jonathan, Rome and Spitzenberg apples in the case of puncture inoculations. The amount of damage caused by Hypochnus rot is probably not large to stored apples in the Northwest, but it seems probable that it is more common than reports would indicate, since it may have been confused with Northwestern anthracnose, perennial canker rot, or Sporotrichum rot because of its very close similarity.

Fusarium Rots

Since Osterwalder (37) studied and described *Fusarium putrefaciens* Ost. producing a decay of apples in Europe in 1904, some fifteen additional species of this genus have been reported as causing a storage or market rot of apple fruit. The attack of most of these species, is incidental and of little importance but at least one species, other than *F. putrefaciens* is outstanding. *F. fructigenum* Fr. is described as one of the five most important species of fungi causing loss from decay of apples in England (16) and in addition, has been reported as the causal agent of bud-rot of apple blossoms (11). Decay caused by species of Fusarium is apparently severe only on market or common storage fruit, since no reports have been found in the literature of the common occurrence of Fusarium rot on cold storage apples.

On cold storage apples from the irrigated sections of eastern Washington, Fusarium species are apparently of very minor importance as causal agents of decay. Cultures of two distinct species or strains of the genus (Fig. 8, A) were infrequently isolated from light brown to tan spots occurring at punctures, and in one instance, one of these forms was obtained from a core rot. Species determination has not been made for these forms. When reinoculated into sound apples, both forms produced decay at cold storage and higher temperatures, and *Fusarium* No. 1 proved to be more virulent than *Fusarium* No. 2. The former produced lesions from 35 to 50 mm. in diameter in 30 days at 68° F., and lesions 15 to 20 mm. in diameter in 60 days at cold storage temperatures. The rotted areas were fairly firm, and light brown to tan color in the centers with usually a darker border, thus giving the "bull's-eye" type of decay. *Fusarium* No. 2 produced lesions 15 to 20 mm. in diameter in 20 days at 68° F., and small, shallow, firm spots at the points of inoculation in 60 days at

cold storage temperatures. The decayed areas were uniformly light brown in color and quite firm. Sporulation was not observed on either species on the fruit, but in the moist atmosphere of the cold storage room, the white mycelium made considerable development on the surface of older lesions.

Ramularia Rot

Three species of Ramularia have been described from apple fruit. *Ramularia macrospora* Fres. was reported from market and storage apples in the United States (7), and in England, *R. magnusiana* (Sacc.) Lind. and *R. heteronema* (B. and Br.) Wollen. (30), have been isolated from apple-rot lesions.

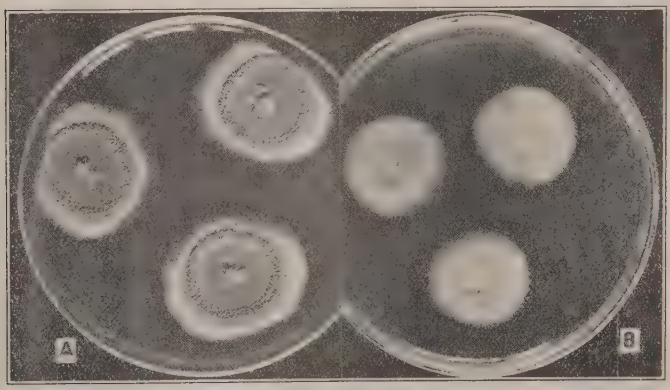

Fig. 10. Isolation cultures on 2 per cent dextrose potato agar. A, **Penicillium expansum**; B, **Ramularia magnusiana.**

Two species of Ramularia have been isolated from decayed spots on Washington apples in cold storage. The first of these is a rapidly growing form in culture, and was isolated but once from a Winesap. It does not agree with any of the forms described from the apple and for the present remains unidentified. The second species (Fig. 10, B) was obtained more frequently and from several varieties and was identified as a strain of *Ramularia magnusiana.* Most of the cultures of this type were isolated late in the storage season, when the apples were very ripe, and there is no indication that injury from Ramularia would ever become serious on cold storage apples.

Ramularia magnusiana produces a slow decay of ripe apples, which is similar in appearance to the rot produced by Fusarium and which develops at about the same rate as the decay produced by our *Fusarium* No. 2. Under moist conditions, it is accompanied by more or less mycelial development on the surface of the lesions. The unidentified Ramularia produces a fairly rapid, light brown, moderately firm rot at 68° F., but at cold storage temperatures is capable of producing small, shallow, dry, firm spots at the points of inoculation, which do not spread to cause active decay. The decay which is formed at 68° F. is sometimes accompanied by sporulation on the surface, the spores being produced in small sporodochia which appear scattered over the surface of the lesion as minute white pustules.

Coryneum Rot

Coryneum foliicolum Fuck. has been reported as capable of causing a slow decay of ripe apples in Maine and is also known to occur in leaf spots and to cause a canker of apple twigs in the orchards of certain sections in eastern United States (32). In England, Horne and Horne (26) record the occurrence of this species on apple fruit associated with small spots and not active rots, and a variety of *C. microstictum* Berk. is also described as causing a rot of apples in that country (30).

Coryneum foliicolum (Fig. 11, A) was isolated in but two instances from small, dark brown, firm lesions which had developed on Rome Beauty

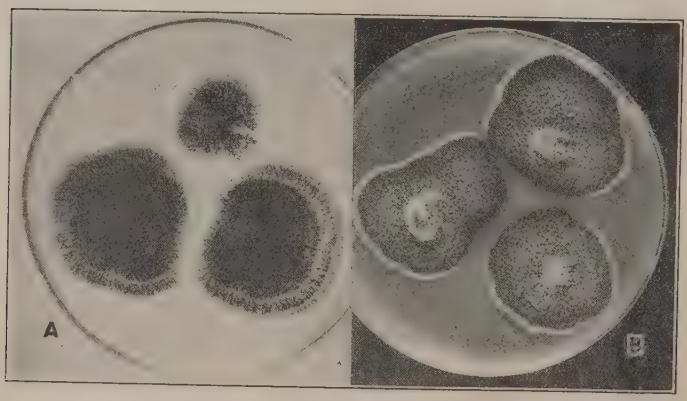

Fig. 11. Isolation cultures on 2 per cent potato agar; A, **Coryneum foliicolum**; B, **Stemphylium congestum**.

apples from the Kennewick District. When inoculated into sound apples, the fungus was found to be weakly parasitic, both at cold storage temperatures and at 77° F.

The lesions produced by Cornyeum are dark brown to almost black, rather shallow, and firm. At cold storage temperatures, the fungus is able to produce small spots about 10 mm. in diameter after several months, but does not spread to produce active decay. At 77° F., the rot progresses slowly and apples will eventually be completely mummified. Sporulation has been observed on the fruit at this temperature, the dark-colored spores being produced in minute, black acervuli beneath the epidermis.

Pestalozzia Rot

Four species of Pestalozzia have been recorded on apple fruit. *Pestalozzia guepini* Desm., *P. brevipes* Cke., and *P. funerea* Desm. have been isolated from market and storage apples in eastern United States (5, 7), and *P. hartigii* Tub., a species known previously only on spruce in Central Europe, was isolated from decayed apples in England (30). A single species of Pestalozzia has been isolated several times from Washington apples in cold storage, and has been identified as *P. hartigii*. When reinoculated into healthy apples, it was found capable of readily parasitizing the fruit, both at cold storage and higher temperatures.

The lesions produced by *P. hartigii* are circular, of a uniform light brown in color, and are fairly firm, with a smooth and slightly sunken surface. The affected host tissue is a light tan in color and is rather dry and spongy. Sporulation has not been observed on the fruit. The decay progresses quite rapidly at 77° F., forming lesions 30 to 35 mm. in diameter in twenty-one days and involving the entire apple in about three months. At cold storage temperature, the decay develops slowly, but much faster than Alternaria rot or other black-rot decay, and eventually apples will be completely destroyed at this temperature.

Pestalozzia rot should probably be classed with the gray molds, since the mycelium of the fungus is white.

Miscellaneous Rots

Several additional fungi have been isolated infrequently from cold storage apples. Because of their rare occurrence and weak parasitism they have been grouped together here.

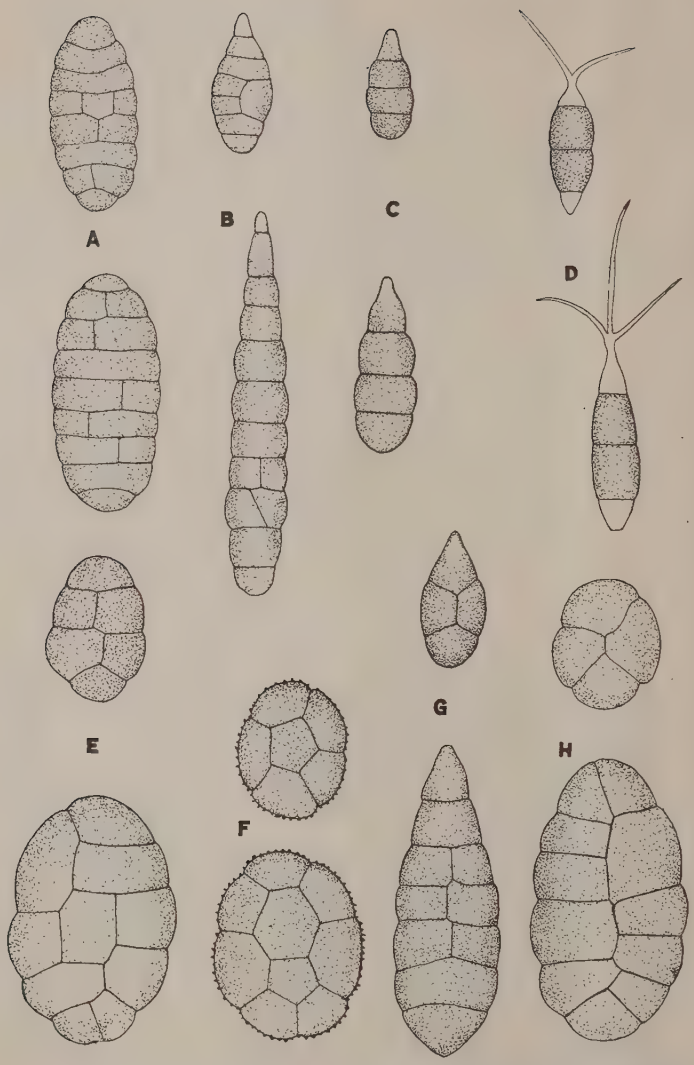

Fig. 12. Drawings of spores of apple-rotting fungi to same scale showing maximum and minimum sizes: A, Ascospores of **Pleospora fructicola**; B, **Alternaria tenuis**; C, **Coryneum foliicolum**; D, **Pestalozzia hartigii**; E, **Pleospora fructicola** (Stemphylium stage); E, **Epicoccum granulatum**; G, **Alternaria mali**; H, **Stemphylium congestum**. All x1000.

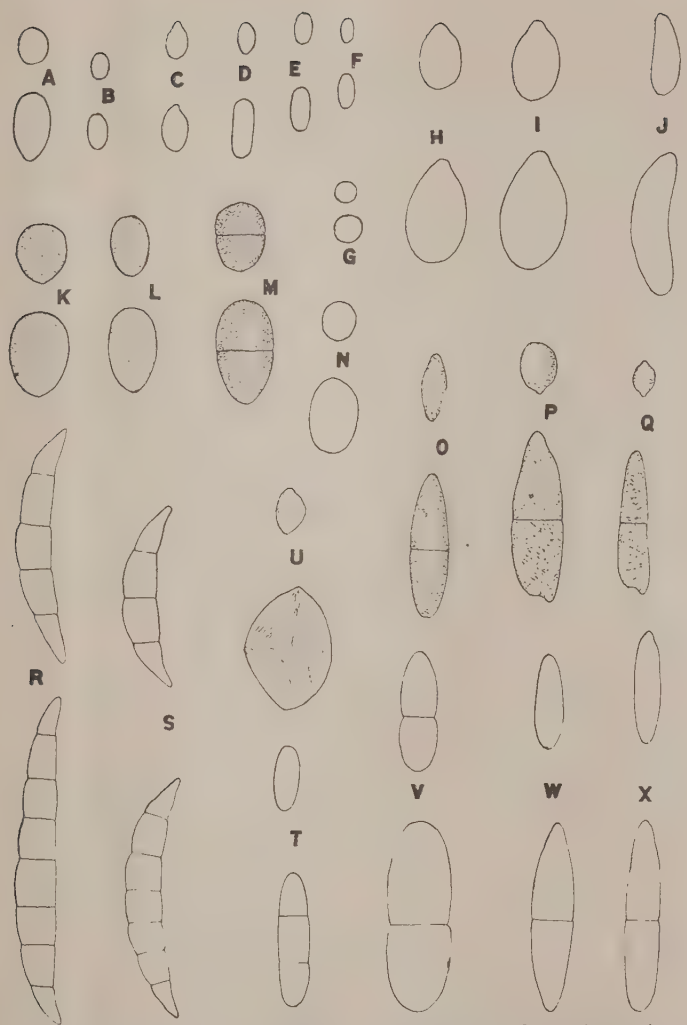

Fig. 13. Drawings of spores of apple-rotting fungi to same scale showing maximum and minimum sizes: A, **Phoma**, No. 2; B, **Phoma** No. 1; C, **Corticium centrifugum**; D, **Sporotrichum malorum**; E, **Cephalosporium carpogenum**; F, **Oospora** spp.; G, **Penicillium expansum**; H, **Botrytis cinerea**; I, **Botrytis mali**; J, **Gloeosporium perennans**; K, **Coniothyrium** No. 2; L, **Coniothyrium** No. 1; M, **Microdiplodia** spp.; N, **Mucor piriformis**; O, **Cladosporium malorum**; P, **Cladosporium herbarum**; Q, **Hormodendron cladosporioides**; R, **Fusarium** No. 2; S, **Fusarium** No. 1 (Macrosonidia); T, **Fusarium** No. 1 (Microconidia); U, **Rhizopus nigricans**; V, Ascospores of **Mycosphaerella tulasnei**; W, **Ramularia** No. 2; X, **Ramularia magnusiana**. All x1000.

A species of Oospora (Fig. 6 B) was isolated once, in association with *Cladosporium herbarum* L., from a small, shallow, dark-colored area bordering a worm hole in a Rome Beauty apple in cold storage. When reinoculated into ripe apples, the fungus was able to produce shallow, brown, firm spots about 10 mm. in diameter after an incubation period of six weeks at 68° F. Pathogenicity has not been determined at cold storage temperatures. The identity of the Washington fungus has not been determined but it seems to be distinct from *Oospora mali* Kidd & Beaum. reported on apple fruit in England (30). A second species of Oospora known as *O. otophila* has recently been reported on this host in Poland (46) but an opportunity for comparison with this species was lacking.

A species of Cephalosporium which has been named *Cephalosporium carpogenum* n. sp. (44) was found to be weakly parasitic on apple fruit. It was isolated in a few instances from small, shallow, brown, firm areas bordering worm holes or punctures. When reinoculated into ripe apples, it was found capable of producing such lesions up to 20 mm. in diameter in two months at 68° F. Pathogenicity has not been determined at cold storage temperatures. A single species of Cephalosporium has been previously reported from apple fruit in England (30) which differs in morphology from *C. carpogenum*.

A species of *Epicoccum*, agreeing closely with the description of *Epicoccum granulatum* Penz. was isolated once from a dark rot on a Rome Beauty apple. It was found in association with the fungus known as *Dematium pullulans*. When inoculated into ripe Jonathan apples, the fungus produced a firm, rather dry, reddish-brown rot, both in cold storage and at 68° F. In cold storage, lesions from 10 to 35 mm. in diameter were formed after five months. At the higher temperature, the rot progressed slowly, forming spots 10 to 12 mm. in diameter in 21 days, which increased to 20 to 25 mm. at the end of two months. In the larger lesions, the affected host tissue was soft, moist and bright red in color, while in the smaller spots the decayed tissue was reddish brown and dry. In some cases cavities were formed beneath the epidermis, which were partially filled with hyphae and fragments of host cells.

METHODS OF INFECTION WITH STORAGE ROTS

The surfaces of normal apples are harboring an innumerable number of the spores of fungi even before they are picked from the trees, but this number may be increased or decreased during the processes of handling up to the packed box (20, 21, 27). Suffice it to say that all packed apples are bearing a load of fungous spores. These may be of species which are unable to cause decay of apple tissue, or they may be forms which are able to digest apple tissue and thus cause decay. These rot-producing species are ever lying in wait for an opportunity to gain an entrance.

There are three general classes of avenues of entrance: (1) Some mechanical injury that has broken the external protective barrier of the thick-walled epidermal layer; (2) necrotic or semi-necrotic surface tissue, that is, groups of cells or more extended areas in which the protoplasmic content has been killed or seriously deranged; (3) the normal unbroken skin of the general surface or accessory parts. Some of the rot-producing fungi are only able to gain an entrance by either the first or second of these avenues, while there are others which can, under certain favorable conditions, infect the perfectly normal fruits.

Special emphasis may be placed upon the ease with which fungi are able to enter through mechanical injuries of various kinds. Although mechanical injuries are supposed to relegate an apple to the culls, certain injuries may escape the sorters. Growth cracks or insect stings or tunnels originate in the orchard, but from the time an apple is first touched by the picker in the orchard until it reposes in the packed box, it must run the gauntlet of nail scratches, pulled stems, stem punctures, box or other cuts, and handling bruises or other man-made injuries.

Observations and experimental tests show that the stem puncture is a wound that is very favorable for infection. With a heavy inoculum of blue mold present it is possible under other favorable conditions to obtain 100 per cent infection, but such high percentages of infection are rarely found in commercial packs. It has been definitely shown that the percentage of infection depends in part upon the number of fungous spores which come in contact with the punctured apples (20, 21). In a recent test of Jonathans through cleaning solutions containing 21-22 blue mold spores per cubic centimeter only 12 of 250 punctures became infected

after three and one-half months in cold storage, while in another lot with an equal number of punctures only one had become infected. Injuries like stem punctures are much more likely to become infected than codling moth stings (See Table 4), as shown by records of culls taken direct from the cull belt and packed for storage. There seemed also to be but little difference in susceptibility to rot of healed and unhealed stings.

Dead tissue is always a source of danger, the chances of infection, however, being reduced if the activity of adjacent cells is sufficient to wall off the killed cells with a barrier of cork. Limb rub lesions, sunscalded areas and skin russeting from frost or sprays are not very likely to be avenues of entrance for rot-producing fungi, unless they are sufficiently severe to cause cracks which penetrate into the flesh. A box of 138 Jonathans with sunscald lesions from the 1930 crop was held in cold storage for three and one-half months without a single spot showing fungous infection. Arsenical, acid or alkali burns from sprays or cleaning solutions may become infected, the percentage of infection sometimes being low, at other times relatively high, consequently injuries of this sort must be guarded against (21). Storage scald offers probably the easiest avenue of entrance through dead or deranged tissue, as many species of decay fungi that can make but little or very slow headway in normal tissue, grow rapidly in the scalded tissue.

There are several avenues through which rot-producing fungi may gain an entrance into normal apples. These are the stems, the calyx, the core, and the lenticels of an unbroken skin. Under certain conditions blue mold may establish itself on the stem and grow down through the stem tissues and finally invade the pulp (3). Excessive humidity and succulent stems appear to favor this type of infection. In light infections of certain fungi which are able to enter through the lenticels, calyx infections sometimes predominate. This has been frequently observed in perennial canker of Yellow Newtown. Either moldy core or a core rot may result if there is an open canal leading from the calyx into the core cells. The seeds may be covered with a mold and the locules filled with the mold mycelium (19), or the fungi may break through the core walls into the surrounding flesh and cause a rot which will advance until it reaches the surface. Open core canals are common in certain varieties, but the percentage of open cores appears to be influenced by the strain and the growing conditions (21). Core rot was observed even in severe form long before the intro-

duction of the practice of washing for removal of spray residue. It has been shown, however, that spray residue removal by immersion is favorable to the inception of core rot, the greatest danger coming from deep and prolonged immersion (21). In these cases the fungous spores are carried into the core, while in ordinary handling the spores probably germinate in the outer portions of the canal and the mycelium may finally grow into the core locules (Fig. 1, B).

While the normal, unbroken epidermis forms an effective barrier against the entrance of many rot-producing fungi, there are other forms that are not excluded, since they are able to penetrate by way of the lenticels. In such cases care in handling will not solve the trouble, but sanitary and protective measures may be of value. Lenticel entrance is the serious phase of such rots as Northwestern anthracnose, perennial canker and the Sporotrichum rot. Recent experience points to the probability that almost any of the rot-producing fungi may under certain favorable conditions enter through the lenticels. It has generally been understood that blue mold can enter only through wounds, via the stems, from the core, or through necrotic tissue, but this does not always hold true, as has been shown by some of our recent records. A lot of Delicious from cold storage examined on January 24, 1931 showed 20 per cent blue mold, and another lot of Winesap showed 25 per cent blue mold, in all cases the infection centering at lenticels. The number of lesions varied from one to a maximum of eight per apple. These observations certainly suggest that lenticel infection may after all be responsible for the high percentage of decay shown by many car lots of apples at eastern terminals (21, 39). Just what the conditions are that must be fulfilled in order to induce lenticel infection are unknown, and present an important problem for further study.

Table 4. Record of the Relation of Certain Cull Defects to the Percentage of Decay Appearing in Storage. 1929 Crop.

Period under observation extended beyond customary commercial storage.

Variety	Defect	No. of apples	No. of blue mold		No. of other rots		Total rots
			Origin from defect listed	Other origin	Origin from defect listed	Other origin	
Delicious	Scald	113	0	6	0	1	7
	Stem puncture	113	16	2	7	0	25
	Healed stings	113	4	5	0	0	9
	Unhealed stings	113	2	2	1	0	5
Spitzenberg	Stem puncture	68	34	2	0	3	39
	Healed stings	54	7	8	0	1	16
Jonathan	No cull	45	1	0	0	0	1
	Unhealed stings	63	2	2	0	0	2
	Healed stings	45	0	2	0	0	2
Rome	Stem puncture	90	46	0	0	7	53
	Healed stings	113	5	2	1	2	10
Winesap	Stem puncture	75	12	0	0	0	12
	Healed stings	164	2	0	0	0	2
	Bitter pit	113	0	4	0	1	5

TEMPERATURE RELATIONS OF STORAGE ROTS

It is not within the scope of the present article to present any detailed discussion of the relation of temperature to the incidence and progress of the different storage rots of apples. Fungi associated with apples may

be grouped into the following classes: (1) Species not causing decay at any temperature; (2) species unable to cause decay in cold storage but capable of causing rapid decay in common storage or on the market; (3) forms causing no decay at cold storage but spot rots at higher temperature; (4) species causing spot rots in cold storage but more rapid decay at higher temperatures; (5) never forming anything but spot rots; and (6) species which may completely rot apples in cold storage during their storage life.

It may be emphasized then, that certain species of the fungi causing decay in cold storage produce only spot rots while others are able to advance sufficiently rapidly to completely rot an apple during its storage life. Only four of the species studied belong to the latter group: Mucor rot (*Mucor piriformis*); blue mold rot (*Penicillium expansum* of the blue mold group); Pestalozzia rot (*Pestalozzia hartigii*) and gray mold rots (*Botrytis* spp.); while all of the others produced lesions of the spot-rot type which grew very slowly, some measuring no more than 7 to 10 mm. (5/16 to ⅜ inches) in surface diameter after four to five months, with the more rapid growers attaining a surface diameter of 35 to 50 mm. (1⅜ to 2 inches) during a similar period. In many cases where spot infections are numerous, the merging of lesions may cause a complete rotting of the fruit, but generally blue mold enters to assist in the destruction.

All of the rots described in the preceding pages occurred in regular cold storage but inoculation tests have brought out one important feature bearing upon the problem of control. Certain species, apparently causally related to the rots from which they were isolated, failed to cause rots when inoculated apples were placed at once in cold storage. This was found in our isolations of *Rhizopus nigricans, Phoma* No. 2 and *Coniothyrium* No. 1. The probable explanation of this behavior is that incipient infections may be established previous to the entrance of fruit into cold storage, and that after once being started they can continue to develop, but initial infection cannot take place at cold storage temperatures. Our experience also points to the fact that some of the rot-producing fungi that are able to start in cold storage, may be greatly favored in their incidence by the holding of apples for some days after harvesting and final packing and storage. In the period between picking and packing two factors may be operating to promote infection: the presence of temperatures favorable to maturing processes in fruit and to the growth

of fungi; and second, the drenching of picked boxes by rains while still standing in the orchard, or a combination of temperature and humidity conditions favorable to infection.

THE PREVENTION OR CONTROL OF STORAGE ROTS

Even with the most careful operations in the orchard, in the packing and storage plants, and in transport to market, it is not possible to entirely eliminate storage rots of apples. Preventive measures are designed to reduce the losses to the lowest possible amount. The measures to which attention should be given may be grouped under three general heads:

1. General sanitation in orchard and packing plant. Since every decay spot originates from a fungous spore that has reached the surface of the apple at some time previous to its final enclosure in the wrap, the general aim of sanitary practices is to reduce the available supply of such spores to the lowest possible point for any given environment. Some of the practices of value may be briefly enumerated:

The control of the orchard phase or stage of rot-producing fungi in cases of known connections, for example, the perennial canker phase of the organism causing one of our bull's-eye rots[1] (*Gloeosporium perennans*).

Removal and destruction of dead limbs, accumulated rubbish and rotting fruit from the orchard.

Use of clean lug boxes or picking boxes rather than old boxes loaded with spores, or sterilization of old boxes if they must be used. Such boxes may be the cause of direct infection or if not they contribute to the constantly increasing supply of fungous spores in the air of the packing room.

Sterilizing of packing rooms and machinery.

Prevention of partially rotted apples from passing into the cleaning solution.

Changing of the cleaning solutions and cleaning the tanks at regular intervals.

Use of fresh or running water rinse, rather than a repump, whenever possible, to wash away as many spores as possible and to reduce danger of arsenical burning.

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[1] Early marketing and consumption of fruit from orchards known to be infested is recommended.

Careful and rigid sanitation in the packing plant including the removal of accumulated rubbish, such as leaves, fruit spurs, and rotting apples, and a reduction of the dust menace to the lowest possible minimum.

Spraying with a fungicide previous to harvesting the fruit has been reported to be of some benefit in perennial canker rot. Sufficient tests are not available to recommend such a practice for general adoption.

2. The avoidance of fruit injuries. Injured or broken skin from insect stings, mechanical breaks in handling, chemical or heat burns, or storage scalding furnish avenues of entrance for rot-producing fungi and consequently all such defects must be reduced to the lowest possible minimum. The following are some of the safeguards:

Have the pickers keep their finger nails clipped short. The use of gloves is sometimes of value.

Do not pinch the fruit in picking and remove with a twisting motion rather than by a pull. Fruit spurs and leaves belong in the orchard, not in the packing plant.

Place the fruit carefully in the picking bucket, but do not drop or throw.

Use a picking bucket that opens at the bottom and in emptying lower carefully into the box so that the apples will roll out gently. Many prefer a picking bucket to a bag, because of less danger of injury to the fruit by bruising.

Be careful in moving and placing ladders and avoid pressing the picking bag against ladders or limbs to avoid bruises.

In order to avoid injury from stacking and loading, do not fill orchard boxes or lugs too full.

Use care in handling and hauling from orchard to packing house.

Use box apron especially with long-stemmed varieties in emptying fruit on the receiving belt of the washer, so that the fruit rolls gently.

Avoid injury by crowding the cleaning machine and grader and thus jamming or stacking the fruit.

Give careful and systematic attention to the mechanical features of all handling machinery, to eliminate sources of injury from rough corners, projecting nails or splinters, unnecessary drops or gravity runs, or hard or unprotected receiving surfaces.

Give careful attention to the cleaning and rinsing to prevent injuries from acid or alkali washes or from arsenical burns.

Guard against too high temperatures of the cleaning solutions. Temperatures up to 110° F. are safe for the time of action and strength of the solutions usually employed. Safety with higher temperatures is uncertain, but temperatures up to 120° F. are sometimes used.

Rigid attention of the sorters to removal of all fruit showing defects that may be avenues of entrance of rot-producing fungi.

Have sorters and packers use gloves to avoid scratches by finger nails and have them clean to avoid scratching by wax nodules, and also because dirty gloves may be rubbing fungus spores into the fruit.

Wrapping the fruit has long been known to reduce losses from storage decay over unwrapped fruit, and the oiled wrap or other substitute for the prevention of scald should always be employed.

The fruit should be packed firmly and tightly but not jammed and too extreme a bulge should be avoided. One to one and one-fourth inches as the sum total of both bulges is very satisfactory. Greater bulges subject the fruit to unfavorable pressure, may cause serious bruising, and in addition appear more favorable to lenticel infection.

Pack so as to avoid stem punctures, that is, with the stem directed into the unoccupied spaces rather than against other fruits.

Special corrugated liners or collars may be used to lessen injury from box pressure and lidding. Two piece lids are preferable to three piece lids unless carefully spaced, and narrow spaces between pieces to wide ones, which may cause lid cuts.

Care in handling the packed boxes, including lidding, movement to storage or in transit to market.

3. The avoidance of conditions which favor or predispose to infection. The features to be emphasized are principally concerned with the direct effect upon the germination of spores and growth of the fungous body, or upon the maturing processes in the fruit. Some of the precautions are:

Avoid delay between picking and packing or movement into cold storage. Ripening processes are speeded up following picking and if fruit is held at orchard temperatures incipient infections are favored.

Avoid delay in picking until the fruit is over mature. Late picking is especially favorable to certain types of spot rots and also to core rot in some varieties.

Avoid allowing boxes of apples to stand in the orchard unprotected during drenching rains. By such a practice two favorable conditions for

fungous infection are supplied: sufficient moisture, and temperatures favorable to spore germination.

Harvesting ahead of the fall rains is of value in lessening certain types of rots, especially those from fungi whose spores depend upon rains for their dissemination.

Avoid cleaning methods in which the fruit is submerged in the cleaning solution or in the rinse because of the danger of carrying spores into the core canal and subsequent development of core rot. Intermittent shallow submersion may be comparatively safe, but long or deep submersion is liable to be disastrous.

Move into storage as quickly as possible and hold the temperature at 30° to 32° for cold storage and as near to that temperature as possible if in common storage. Some fungi can not produce infections at cold storage temperatures, while others are very greatly retarded in their progress.

If picked fruit must be held for some days before packing, losses may be prevented or lessened by holding in cold storage and packing as soon as possible. (Holding for a long time before packing is sometimes disastrous).

Use care in stacking large quantities of packed boxes in rapid succession so as to permit as rapid cooling as possible, because high close stacks retard the equalization of temperatures.

Provide, if possible, for a controlled humidity of the storage rooms. Too low a humidity results in losses from shrivelling, while too high a humidity is favorable for fungous infection. An humidity of 83 to 85 per cent is recommended for cold storage.

Some tests and observations have indicated a possible relation between soil types or soil fertility and susceptibility to decay, but available data are not sufficiently complete to justify definite statements.

SUMMARY

1. The importance of the apple industry has been emphasized, followed by a classification of fruit defects and a discussion of the importance of decay of apples in storage or during transit to market.

2. A study is presented of the types of apple rots in Washington, the forms being arranged in the order of their importance and according to the generic relationship of the causal organisms.

3. More than 40 species of fungi belonging to 22 genera are recorded as connected with the decay of apples in cold storage in Washington. Two belong to the Phycomycetes, two to the Ascomycetes, one to the Basidiomycetes, but all the others to the Fungi Imperfecti.

4. Several of the most destructive fungi such as blue mold may cause a complete decay of fruits during their cold storage life, but the majority are slow growing at cold storage temperatures and produce spot rots, several of which are of the bull's-eye type, that is, show a light center, with a darker border.

5. Three general avenues of entrance of rot-producing fungi are discussed: (1) Mechanical injuries; (2) necrotic or semi-necrotic surface spots or tissue; and (3) the normal, unbroken skin.

6. The importance of the temperature factor in the onset and progress of apple-rotting fungi is emphasized.

7. Recommendations for the prevention or control of storage rots of apples are grouped under three general heads: (1) General sanitation in the orchard and packing plant; (2) the avoidance of injuries to the fruit which permit entrance of rot-producing fungi; and (3) the avoidance of conditions which favor or predispose to infection.

LITERATURE CITED

1. Adam, D. B., Experiments in the storage of fruits. Jour. Dept. Agr. Victoria **21**: 371-382. 1923.
2. Anderson, P. J. et al., Check list of diseases of economic plants in the United States. U. S. Dept. Agr. Bul. **1366**: 1-111. 1926.
3. Barnum, C. C., Stem-end rot of apples. Science N. S. **55**: 707-708. 1922.
4. Behrens, J., Beiträge zur Kentnis der Obstfäulnis. Centrabl. f. Bakt., II Abt. **4**. 514-522, 547-553, 577-585, 635-644, 700-745, 770-777. 1898.
5. Brooks, E. and Cooley, J. S., Temperature relations of apple rot fungi. Jour. Agr. Res. **8**: 139-163. 1917.
6. Brooks, C., Cooley, J. S. and Fisher, D. F., Diseases of apples in storage. U. S. Dept. Agr., Farmers' Bul. **1160**: 1-23. 1920.
7. Brooks, Charles; Fisher, D. F. and Cooley, J. S., Apple rots. Abstr. in Phytopath. **4**: 403. 1914.
8. Butler, L. F., Increasing prevalence of Hypochnus rot of apples. Phytopath. **17**: 734-744. 1927.
9. Butler, L. F., Corticium centrifugum, a heterothallic pathogene of apples. Jour. Agr. Res. **41**: 269-294. 1930.
10. Cunningham, G. H., Delicious spot. In Fungous Diseases of Fruit Trees, p. 185. Brett Printing and Publ. Co., Auckland. 1925.
11. Dillon-Weston, W. A. R., A note on the bud rot of apple trees. Trans. Brit. Myc. Soc. **12**: 170-172. 1927.
12. Eustace, H. J., Two decays of stored apples. New York State Agr. Exp. Sta. Bul. **297**: 31-48. 1903.
13. Fisher, D. F., New apple rot unmasked and named. Better Fruit, **21**: 9. 1926.
14. Fisher, D. F., A new apple rot disease, bull's-eye rot, caused by Gloeosporium perennans. Proc. Wash. State Hort. Assoc. **21**: 44-48. 1926.
15. Fisher, D. F., and Reeves, E. L., Perennial canker. Proc. Wash. State Hort. Assoc. **24**: 55-61. 1928.
16. Fruit and Vegetable Committee, Section V. Rept. Food Invest. Board for 1923. Dept. Sci. and Ind. Res. 1923: 33-65. 1924.
17. Gardner, M. W., Sporotrichum fruit spot of apple. Abstr. in Phytopath. **18**: 145. 1928.
18. Gardner, M. W., Sporotrichum fruit spot and surface rot of apple. Phytopath. **19**: 443-452. 1929.
19. Heald, F. D., Moldy core of Stayman Winesap. Abstr. in Phytopath. **11**: 105. 1921.
20. Heald, F. D., Blue mold in relation to fruit cleaning methods. Proc. Wash. State Hort. Assoc. **23**: 143-148. 1927.
21. Heald, F. D., Neller, J. R. and Overley, F. L., Arsenical spray residue and its removal from apples and pears. Wash. Agr. Exp. Sta. Bul. **226**: 1-100. 1928.
22. Heald, F. D., and Sprague, Roderick., A spot rot of apples in storage caused by Botrytis. Phytopath. **16**: 485-488. 1926.
23. Hesler, L. R., The perfect stage of *Hendersonia mali*. Mycologia **19**: 222-227. 1927.
24. Horne, A. S., Diagnosis of fungi from "spotted" apples. Jour. Bot. (London) **58**: 238-242. 1920.
25. Horne, A. S. and Horne, E. V., On spotting of apples. Gardner's Chronicle **68**: 216. 1920.
26. Horne, A. S. and Horne, E. V., On the spotting of apples in Great Britain. Ann. Appl. Biol. **7**: 183-201. 1920.

27. Huber, Glenn A., The Aspergilli and their relation to decay in apples. Jour. Agr. Res. **41**: 801-817. 1930.
28. Ivanoff, B., Diseases of pome fruits and measures for their control. Abstr. in Rev. Appl. Myc. **4**: 418-419. 1925.
29. Janczewski, E., Recherches sur le *Cladosporium herbarum* et ses compagnons habituels sur le cerales. Bul. Acad. Science Cracovie **27**: 187-207. 1894.
30. Kidd, M. N., and Beaumont, A., Apple rot fungi in storage. Trans. Brit. Mycol. Soc. **10**: 98-118. 1924.
31. Kidd, M. N., and Beaumont, A., An experimental study of the fungal invasion of apples in storage with particular reference to invasion through lenticels. Ann. Appl. Biol. **12**: 14-33. 1925.
32. Lewis, C. E., Apple disease caused by *Coryneum foliicolum* and *Phoma mali*. Maine Agr. Exp. Sta. Bul. **170**: 185-200. 1909.
33. Mac-Innes, F. J. The occurrence of Alternaria in a characteristic apple spot, and an apple rot caused by *Gliocladium viride*. Trans. Ill. Acad. Sci. **10**: 218-223. 1917.
34. Morse, W. J., and Lewis, C. E., Maine apple diseases. Maine Agr. Exp. Sta. Bul. **185**: 337-392. 1910.
35. Newton, G. A., Some fungi of the Stemphylium type and their relation to apple rots. Phytopath. **18**: 565-578. 1928.
36. O'Gara, P. J., The parasitism of *Coniothyrium fuckelii*. Phytopath. **1**: 100-102. 1911.
37. Osterwalder, A., Ueber eine bisher unbekannte Art der Kenobstfäule verursacht durch Fusarium putrefaciens nov. spec. Centralb. f. Bakt. II Abt., **13**: 207-213; 330-338. 1904.
38. Oudemans, C. A. J. A., Enumeratio Systematica Fungorum. Vol. 3. Martinus Nijhoff, The Hague. 1919.
39. Pailthorp, R. R., and Park, J. W., Extent and causes of rejections of boxed apples from the State of Washington, Season of 1922 to 1925. U. S. Dept. Agr. Circ. **413**: 1-15. 1927.
40. Roberts, J. W., Experiments with leaf-spot fungi. Jour. Agr. Res. **11**: 57-65. 1914.
41. Roberts, J. W., Morphological characters of *Alternaria mali* Roberts. Jour. Agr. Res. **27**: 699-708. 1924.
41a. Rose, D. H., and Butler, L. F., Pleospora rot of lemons and apples. Abstr. in Phytopath. **17**: 47. 1927.
42. Ruehle, G. D., *Stemphylium congestum* and its relation to decay in apples. Mycologia **22**: 304-309. 1930.
43. Ruehle, G. D., Cladosporium species from apple fruit and the perfect stage of Cladosporium herbarum. Abstr. in Phytopath. **20**: 854. 1930.
44. Ruehle, G. D., New species of apple rot fungi from Washington. Phytopath. **21**: (In press). 1931.
45. Schneider-Orelli, O., Versuche ueber die Wachstumsbedingungen und Verbreitung der Fäulnispilze des Lagerobstes. Centralbl. f. Bakt., II Abt., **32**: 161-169. 1912.
46. Siemaszko, W., Notaki fitopatologiczne I. Reprinted from Choroby i Szkodniki Roslin **1**: 1-4. 1925. Abstr. Rev. Appl. Myc. **4**: 380. 1925.
47. Thom, Charles, The Penicillia, pp. 1-643. Williams and Wilkins Co., Baltimore, Md. 1930.
48. Zeller, S. M., and Childs, L., Perennial canker of apple trees. Oregon Agr. Exp. Sta. Bul. **217**: 1-17. 1925.